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A39

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification ⁷ : A61K 38/24, 38/04, 9/14, 9/52	A1	(11) International Publication Number: WO 00/40259 (43) International Publication Date: 13 July 2000 (13.07.00)
(21) International Application Number: PCT/KR99/00071		(81) Designated States: BR, CA, CN, IN, JP, MX, TR, US, European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE).
(22) International Filing Date: 11 February 1999 (11.02.99)		
(30) Priority Data: 1998/62142 30 December 1998 (30.12.98) KR		Published <i>With international search report.</i>
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(54) Title: PROLONGED RELEASE MICROSPHERE ENCAPSULATING LUTEINIZING HORMONE-RELEASING HORMONE ANALOGUES AND METHOD FOR PREPARING THE SAME		
(57) Abstract		<p>There is disclosed a prolonged release microsphere which can constantly release medicinal drugs, such as luteinizing hormone-releasing hormone analogues and encapsulate them at high content rates. It is prepared by dissolving a copolymer of lactide and glycolide in methylene chloride, dissolving a luteinizing hormone-releasing hormone analogue and a release-controlling material in a subsidiary solvent, combining the above two solutions with each other to produce an emulsion phase, dispersing the emulsion phase in a solution of polyvinyl alcohol in distilled water to give a single emulsion system, removing the combined solvent of the emulsion phase to generate a polymeric microsphere; freeze-drying the polymeric microsphere. The microsphere prepared has a much finer inner structure, by virtue of which the microsphere is secured in a constant release rate. The single emulsion system which simplifies the preparation, allows for the maintenance of a drug content of 10 % or more. The charged groups of the release-controlling materials associated with the polymers minimize the excess release of the oppositely charged drugs at an initial stage, playing an important role in keeping the release rate constant.</p>

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PROLONGED RELEASE MICROSPHERE ENCAPSULATING LUTEINIZING HORMONE-RELEASING HORMONE ANALOGUES AND METHOD FOR PREPARING THE SAME

Technical Field

5 The present invention relates to a microsphere encapsulating luteinizing hormone-releasing hormone (hereinafter referred to as "LHRH") analogues, which is able to constantly release them for a long period of time. Also, the present invention is concerned with a method for preparing such a prolonged release microsphere.

10 Background Art

15 Physiologically, when testosterone or estrogen is in a low concentration level in blood or when hypothalamic-releasing hormone is stimulated, gonadotropin-releasing hormone (hereinafter referred to as "GnRH") is secreted from the hypothalamus. The GnRH is then transferred through the hypothalamic-pituitary portal system to the pituitary gland at which the GnRH stimulates the synthesis and secretion of luteinizing hormone (hereinafter referred to as "LH") and follicle stimulating hormone. As a result, testosterone or estrogen is secreted.

20 LHRH analogues act on the pituitary gland to inhibit the secretion of LH, thus resulting in the antagonizing of the liberation of testosterone and estrogen into the bloodstream. By taking advantage of this antagonistic action, the diseases caused by testosterone and estrogen, such as prostatic cancer, breast cancer, endometriosis and the like, have recently been therapeutically treated.

25 Like general peptide drugs, LHRH is, however, very unstable within the gastro-intestinal tract and shows a low uptake efficiency therein. Therefore, the administration of LHRH has been usually performed via injection. The administration via injection also has a significant disadvantage of being very poor in bioavailability so that LHRH is required to be injected daily. Such injection administration also requires a long cure period, which causes a problem in a patient's adaptation to the drug, therapeutic efficiency, and treatment.

30 Extensive research has been made on the use of poly(lactide-co-

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glycolide) (PLGA), which is an intravital degradable polymer, in controlled releasable dosage forms which contain proteins or peptides. The released form and structural stability of proteins with high molecular weights are the most difficult barriers in commercializing them into medicinal drugs. In result, research efforts have been and continue to be directed to the development of various additives and new preparation processes by which the proteins can be commercialized while maintaining their activity.

For peptide drugs, commercialization is relatively easy because peptides are smaller in size and stabler than proteins. As commercialized examples, there are sustained-release DOS preparations containing LHRH analogues, which are peptides with a molecular weight of 1.2 kDa.

The microspheres made of PLGA are decomposed into lactic acid and glycolic acid by hydrolysis in the body and metabolized. In other words, PLGA is not harmful to the human body nor shows side effects from the decomposed products. What is better, the microspheres made of PLGA can release their contents, e.g. therapeutically effective ingredients, at a constant rate for a desired period of time.

In this regard, U.S. Pat. No. 4,711,782 introduces a technique for producing microporous microcapsules from similar polymers by W/O/W (water in oil in water) double emulsification. This technique is usually used to capsulate water-soluble drugs. In this technique, when a water-soluble drug is dissolved in water, gelatin is used together to retain the drug, and this aqueous layer is dispersed in an organic layer containing the polymer with the aid of a homogenizer, so as to give a primary emulsion. Again, this primary emulsion is dispersed in water containing polyvinyl alcohol as a surfactant, to give a secondary emulsion. The organic solvent is diffused into the aqueous layer and evaporated, so that the polymer is solidified to form the microcapsules. They are then freeze-dried.

As mentioned, water is used to dissolve water-soluble peptides and the microcapsules obtained are of porous structure. Thus, these microcapsules have a problem of being so high in the initial release rate of peptide drugs and low in drug content.

Disclosure of the Invention

The intensive and thorough research on a prolonged release microsphere, repeated by the inventors aiming to release peptide drugs continuously for an extended period of time, resulted in the finding that, 5 when an appropriate combination of two poly(lactide-co-glycolide) copolymers, which have an equimolar ratio between their lactide moiety and glycolide moiety and have a carboxyl group and a dodecyl group at their ends, is used as a carrier for drug, the microsphere is enhanced in biodegradation rate as well as in drug content. The negative charge of the 10 carboxyl group attached to the end of the biodegradable polymer form an ion bond with the positive charges that the peptide drugs possess, increasing the drug content in the microsphere and preventing the drugs from being released excessively at an initial time due to diffusion. The dodecyl group plays an important role in controlling the degradation rate 15 of the microsphere. Consequently, the microspheres in the body release LHRH analogues continuously to maintain the concentration of testosterone and estrogen in blood for an extended period of time, so as to improve the therapeutic efficiency of and the patient's adaptation to the drugs.

20 Therefore, it is an object of the present invention to provide a prolonged release microsphere which can control the release of drugs for a sustained period of time.

25 It is another object of the present invention to provide a prolonged release microsphere which is high in the content of therapeutically effective ingredients.

It is a further object of the present invention to provide a method 30 for preparing such a prolonged release microsphere with ease and a good efficiency.

In accordance with an aspect of the present invention, there is 35 provided a prolonged released microsphere, which is composed of a poly(lactide-co-glycolide) copolymer and encapsulate a luteinizing hormone-releasing hormone analogue.

In accordance with another aspect of the present invention, there is provided a method for preparing a prolonged release microsphere, comprising the steps of: dissolving a copolymer of lactide and glycolide in methylene chloride; dissolving a luteinizing hormone-releasing

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hormone analogue and a release-controlling material in a subsidiary solvent; combining the above two solutions with each other to produce an emulsion phase; dispersing the emulsion phase in a solution of polyvinyl alcohol in distilled water to give a single emulsion system; removing the combined solvent of the emulsion phase to generate a polymeric microsphere; and freeze-drying the polymeric microsphere.

Best Modes for Carrying Out the Invention

In the present invention, a microsphere which retains therapeutically effective drugs and continuously releases them for a sustained period of time, is prepared from a mixture of biodegradable polymers. As therapeutically effective drugs, LHRH analogues are of particular interest. Therefore, if the microspheres retaining LHRH analogues are administered, they release the drugs for a long period of time, whereby testosterone or estrogen can be maintained at a low level in the blood and an improvement in therapeutic effect and patient's adaptation to the drugs can be brought about.

The prolonged release microsphere which encapsulates LHRH analogues has an aporous, fine and uniform inner structure, so that its drug content rate is enhanced.

The microsphere can be prepared in a single emulsion process. In detail, LHRH analogues, including goserelin acetate, nafarelin acetate, buserelin acetate and leuprorelin acetate, are dissolved in a subsidiary solvent and added to an organic solvent containing a polymer to give an oil phase which is then dispersed in an aqueous phase.

A preferred example of the organic solvent useful in the invention is methylene chloride. The aqueous phase is obtainable by dissolving polyvinyl alcohol.

The subsidiary solvent which dissolves LHRH should be miscible with the organic solvent (methylene chloride) and water, both. Examples of the subsidiary solvent include N-methyl-2-pyrrolidone (NMP), dimethyl sulfoxide (DMSO), dimethyl formamide (DMF), acetone, ethanol, ethyl acetate, and methyl ethyl ketone (MEK) with the most preference to NMP. This subsidiary solvent plays an important role in providing the microsphere with an aporous fine structure.

In the present invention, the release of LHRH analogues is

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controlled in a double manner by the actions of the functional groups, i.e. carboxyl group and dodecyl group, attached to the ends of the two polymers which compose the microsphere. The carboxyl group forms a hydrophobic ion pair with LHRH analogues, so the release rate thereof is retarded. The dodecyl group inhibits the enzymatic action to degrade the microsphere, so the integrity of the biodegradable microsphere is sustained. Therefore, the drug contained in the microsphere is not released in a sudden burst.

The compound suitable to retard the release rate of LHRH analogues must form the hydrophobic ion pair with LHRH analogues as well as can be dissolved in the organic solvent. Preferable examples to meet these standards include sodium oleate, deoxycholic acid, cholic acid, fatty acids and phosphatidic acids.

The biodegradable microsphere of the present invention is aporous with an ultrafine inner structure, as shown in Figs. 1 and 2. The data obtained from an *in vitro* release test demonstrate that LHRH is released at relatively constant rates from the microspheres of the invention, as shown in Fig. 3. The microspheres were measured for their weight loss in order to obtain the information about their biodegradation rates, which finally told that the microspheres are completely decomposed on around the 45th day after testing, as shown in Fig. 4. The data obtained from an *in vivo* release test, shown in Fig. 5, are well correlated with those of Fig. 4.

A better understanding of the present invention may be obtained in light of the following examples which are set forth to illustrate, but are not to be construed to limit the present invention.

EXAMPLE I : Preparation of Biodegradable Microsphere Containing Leuprorelin acetate

A microsphere was made from biodegradable PLGA in an O/W (oil in water) mono-emulsification method.

In 3 ml of methylene chloride was dissolved 350 mg of each of a PLGA which has a dodecyl group at its end and a molecular weight of 12,000 with 50:50 lactide moiety:glycolide moiety, such as that sold by Boehringer Ingelheim under the brand name of RH502, and a PLGA which has a carboxyl group at its end and a molecular weight of 8,600

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with 50:50 lactide moiety:glycolide moiety, such as that sold by Boehringer Ingelheim under the brand name of RH502H. This methylene chloride solution was sufficiently mixed with a solution of 100 mg of leuprorelin acetate in 1 ml of N-methyl-2-pyrrolidone. To the saturated solution with the polymer and the drug, 250 ml of a solution of 0.5 wt% polyvinyl alcohol in distilled water containing 2 g of methylene chloride was added and, then, emulsified by using a stirrer at 700 rpm for 30 min. While the emulsion was stirred for 3 hours under an atmosphere, N-methyl-2-pyrrolidone was extracted with water and methylene chloride was evaporated out, so as to give solidified microspheres. After being collected by centrifugation at 8,000 rpm for 30 min, the microspheres were centrifuged again twice with water to remove remaining solvent and drugs and then, freeze-dried.

EXAMPLE II : Preparation of Biodegradable Microsphere Containing
Goserelin acetate

In 3 ml of methylene chloride were dissolved 100 mg of each of RG502H and RG502. This methylene chloride solution was sufficiently mixed with a solution of 25 mg of leuprorelin acetate in 1 ml of N-methyl-2-pyrrolidone. To the saturated solution with the polymer and the drug, 200 ml of a solution of 0.3 wt% polyvinyl alcohol in distilled water containing 2 g of methylene chloride was added and, then, emulsified by using a stirrer at 700 rpm for 30 min. Thereafter, microspheres were prepared by following the remaining procedure of Example I.

EXAMPLE III: Preparation of Biodegradable Microsphere Containing
Nafarelin acetate

In 5 ml of methylene chloride were dissolved 200 mg of each of RG502H and RG502. This methylene chloride solution was sufficiently mixed with a solution of 50 mg of nafarelin acetate in 1 ml of N-methyl-2-pyrrolidone. To the saturated solution with the polymer and the drug, 250 ml of a solution of 0.3 wt% polyvinyl alcohol in distilled water containing 2 g of methylene chloride was added and, then, emulsified by using a stirrer at 500 rpm for 30 min. Thereafter, microspheres were prepared by following the remaining procedure of Example I.

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EXAMPLE IV: Preparation of Biodegradable Microsphere Containing Leuprorelin Acetate using Homogenizer

In 5 ml of methylene chloride were dissolved 200 mg of each of RG502H and RG502. This methylene chloride solution was sufficiently mixed with a solution of 50 mg of leuprorelin acetate in 1 ml of N-methyl-2-pyrrolidone. To the saturated solution with the polymer and the drug, 250 ml of a solution of 0.5 wt% polyvinyl alcohol in distilled water containing 2 g of methylene chloride was added and, then, emulsified by using a homogenizer at 700 rpm for 30 min. Thereafter, microspheres were prepared by following the remaining procedure of Example I.

EXAMPLE V: Preparation of Biodegradable Microsphere Containing Leuprorelin acetate with Sodium Oleate

In 1 ml of methylene chloride were dissolved 200 mg of each of RG502H and RG502. This methylene chloride solution was sufficiently mixed with a solution of 50 mg of leuprorelin acetate and 3.105 mg of sodium oleate in 1 ml of N-methyl-2-pyrrolidone. To the saturated solution with the polymer and the drug, 250 ml of a solution of 0.3 wt% polyvinyl alcohol in distilled water containing 2 g of methylene chloride was added and, then, emulsified by using a homogenizer at 700 rpm for 30 min. Thereafter, microspheres were prepared by following the remaining procedure of Example I.

EXAMPLE VI: Preparation of Biodegradable Microsphere Containing Sodium Oleate/Leuprorelin Complex

17.5 mg of sodium oleate and 50 mg of leuprorelin acetate were reacted in distilled water to yield precipitates which were, then, collected and freeze-dried. They were dissolved in a mixed solution of 0.66 ml of N-methyl-2-pyrrolidone and 1.33 ml of methylene chloride which contained 200 mg of each of RG502H and RG502. To the saturated solution with the polymer and the drug, 250 ml of a solution of 0.3 wt% polyvinyl alcohol in distilled water containing 2 g of methylene chloride was added and, then, emulsified by using a homogenizer at 700 rpm for 30 min. Thereafter, microspheres were prepared by following the

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remaining procedure of Example I.

TABLE 1
Drug Content and Average Particle Size of the Microsphere

Examples	% Drug Content	Avg. Particle Size (μm)
I	10.76	138.5
II	9.98	106.8
III	10.21	122.1
IV	10.71	11.2
V	10.32	10.6
VI	11.21	10.4

TEST EXAMPLE I: *in vitro* Drug Release of Microspheres

The biodegradable microspheres prepared in Examples were tested for *in vitro* release as follows. 5 mg of the freeze-dried microsphere were dispersed in a vial containing a solution of 0.05% Tween 80 in 10 ml of a 0.333 M phosphate buffer and stored at 37 °C for 28 days. A test sample was taken every third day from the first day to the thirtieth day. The ten samples thus taken were centrifuged. After the removal of the supernatant, the microspheres were quantified for the drugs through an HPLC with a mobile phase of 3:1 ammonium acetate:methanol at a flow rate of 1.0 ml/min at 280 nm. The results were shown in Fig. 3.

TEST EXAMPLE II: Degradation of Microspheres

Under the same condition as that of Test Example I, a test sample was taken every forth day. The samples were centrifuged, followed by the removal of the supernatant. The microspheres thus obtained were dried and accurately measured for their weights. From the measurements, the degradation rates of the microspheres were calculated, and shown in Fig. 4.

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TEST EXAMPLE III: *in vivo* Drug Release of Microspheres

The biodegradable microspheres prepared in Examples were tested for *in vitro* release as follows. The microspheres were introduced into the femoral regions of rats via intramuscular injection and the remaining microspheres were taken from the femoral regions by incising the regions every fifth day. The microspheres taken were homogenized in 10 ml of a solution of 0.02 wt% Tween 80 (polyoxyethylene 20 oleate, Junsei Chemical Co.) in a 0.333 M phosphate buffer (pH 7.0). After further addition of 10 ml of the buffer and 10 ml of methylene chloride, the drugs were extracted in an aqueous layer. These extracts were quantified by HPLC under the same condition as that of the *in vitro* release test and the results are shown in Fig. 5.

Brief Description of the Drawings

Fig. 1 is an SEM photograph showing the microsphere of the present invention;

Fig. 2 is an SEM photograph showing a cross section of the microsphere of the present invention.

Fig. 3 is a plot showing the *in vitro* release rates of the microspheres against time.

Fig. 4 is a plot showing the weight loss rates of the microspheres against time.

Fig. 5 is a plot showing the *in vivo* release rates of the microspheres against time.

Industrial Applicability

As described hereinbefore, the microspheres prepared according to the present invention have much finer inner structures than do conventional microspheres, by virtue of which the microspheres are secure in a constant release rate. The single emulsion system of the present invention simplifies the preparation process of the microsphere, enabling it to maintain a drug content of 10% or more. In addition, the charged groups of the release-controlling materials associated with the polymers minimize the excess release of the oppositely charged drugs at

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an initial stage, playing an important role in keeping the release rate constant.

The present invention has been described in an illustrative manner, and it is to be understood the terminology used is intended to be in the nature of description rather than of limitation. Many modifications and variations of the present invention are possible in light of the above teachings. Therefore, it is to be understood that within the scope of the appended claims, the invention may be practiced otherwise than as specifically described.

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CLAIMS

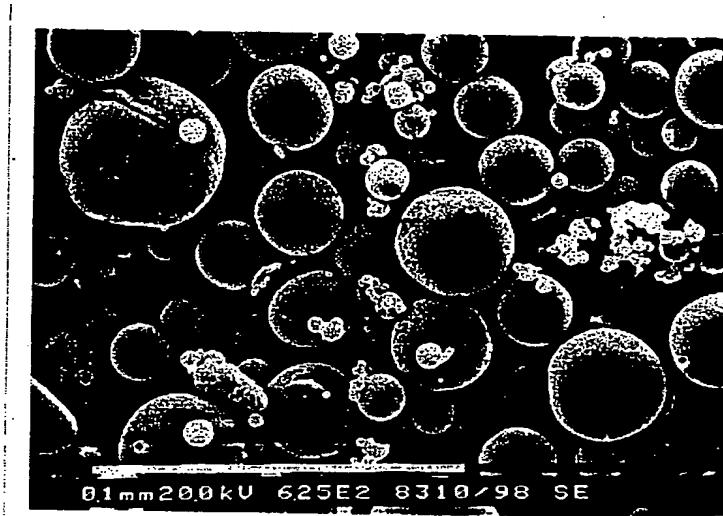
1. A prolonged released microsphere, which is composed of a poly(lactide-co-glycolide) copolymer and encapsulate a luteinizing hormone-releasing hormone analogue.
- 5 2. A prolonged released microsphere as set forth in claim 1, wherein the luteinizing hormone-releasing hormone analogue is selected from the groups consisting of goserelin acetate, nafarelin acetate, buserelin acetate and leuprorelin acetate.
- 10 3. A prolonged released microsphere as set forth in claim 1, wherein the copolymer consists of a polylactide and a polyglycolide either of which have a dodecyl group and a carboxyl group at their ends.
- 15 4. A method for preparing a prolonged release microsphere, comprising the steps of:
 - dissolving a copolymer of lactide and glycolide in methylene chloride;
 - dissolving a luteinizing hormone-releasing hormone analogue and a release-controlling material in a subsidiary solvent;
 - 20 combining the above two solutions with each other to produce an emulsion phase;
 - dispersing the emulsion phase in a solution of polyvinyl alcohol in distilled water to give a single emulsion system;
 - removing the combined solvent of the emulsion phase to generate a polymeric microsphere;
 - 25 freeze-drying the polymeric microsphere.
- 25 5. A method as set forth in claim 4, wherein the single emulsion system comprises 75.0-99.0 wt% of an aqueous phase and 0.3-0.5 wt% of polyvinyl alcohol and the emulsion phase comprises 0.50-10.0 wt% of methylene chloride and 0.2-10.0 wt% of the subsidiary solvent.
- 30 6. A method as set forth in claim 4 or 5, wherein the subsidiary solvent is N-methyl-2-pyrrolidine.

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7. A method as set forth in claim 5, wherein the release-controlling material is capable of hydrophobic ion paring with the luteinizing hormone-releasing hormone analogue and being dissolved in an organic solvent.
- 5 8. A method as set forth in claim 5 or 7, wherein the release-controlling material is sodium oleate and is used at an amount of 75-100 mol% based on the moles of the luteinizing hormone-releasing hormone analogue.

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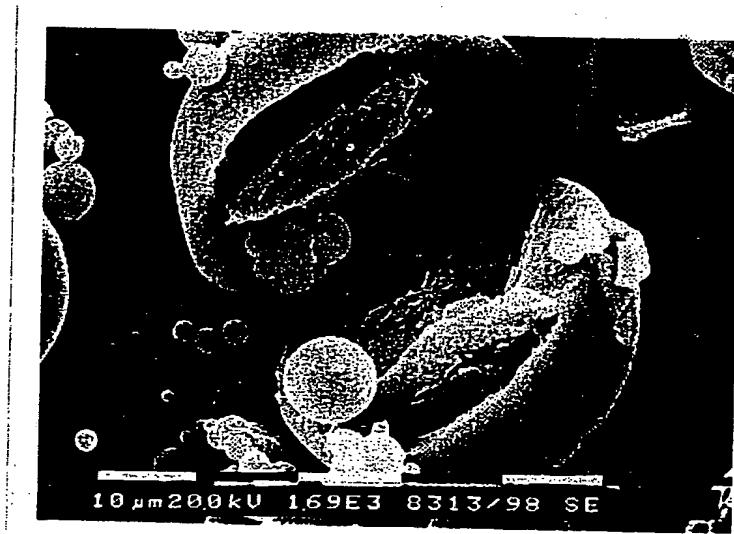
FIG. 1



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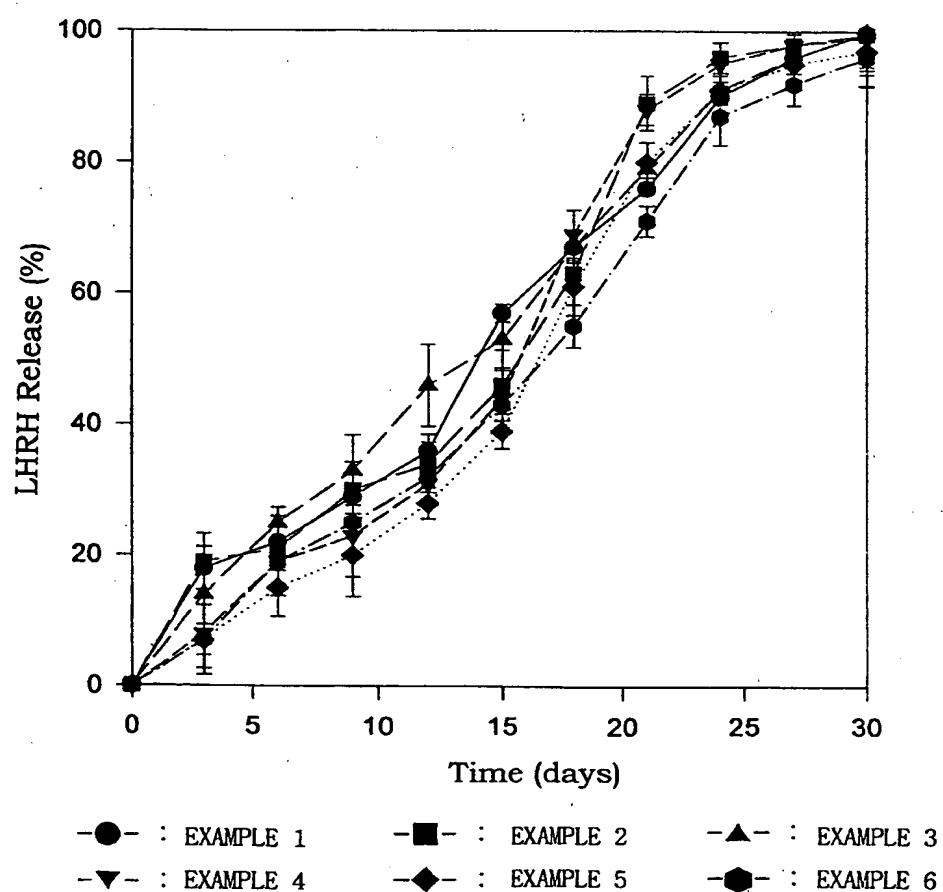
FIG. 2



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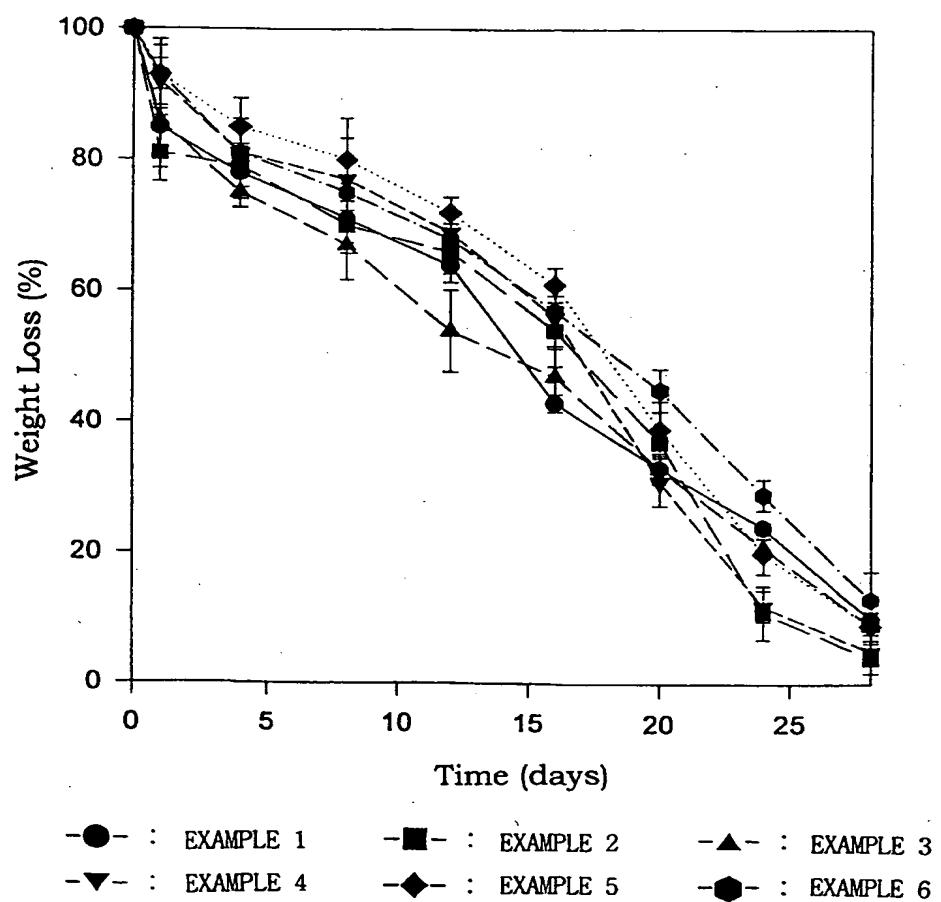
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FIG.3



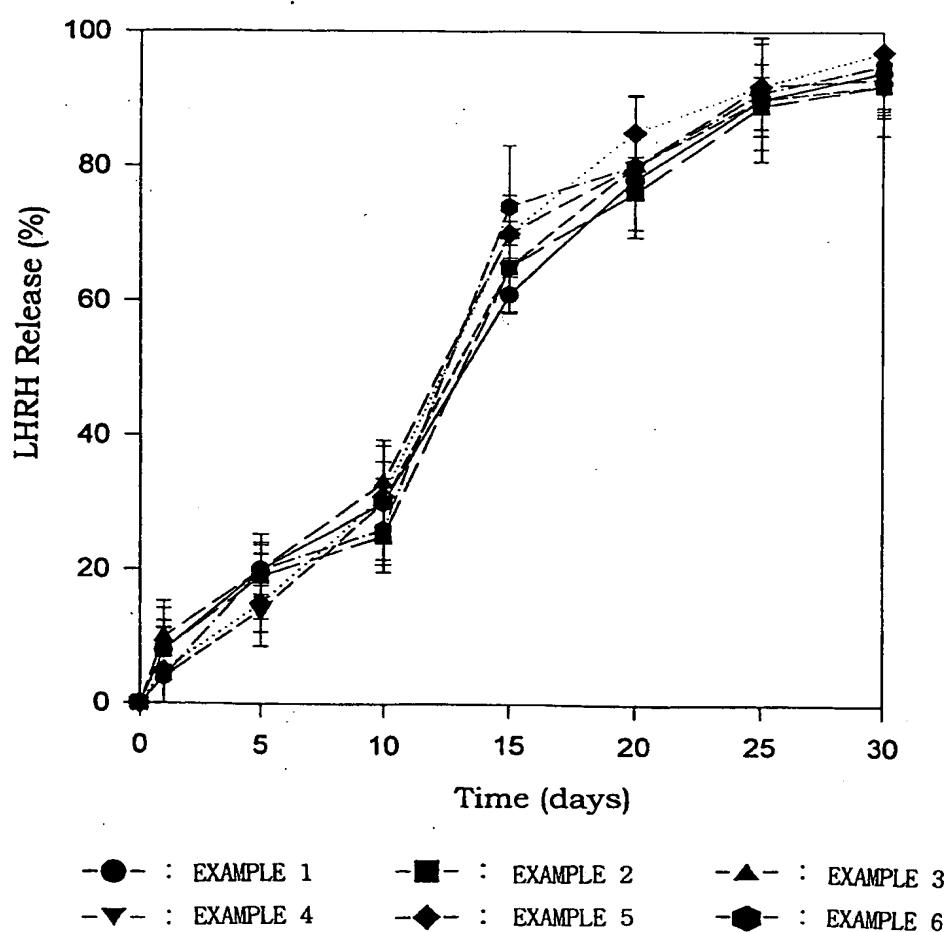
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FIG.4



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FIG.5



INTERNATIONAL SEARCH REPORT

International application No.
PCT/KR 99/00071

A. CLASSIFICATION OF SUBJECT MATTER

IPC⁷: A 61 K 38/24, 38/04, 9/14, 9/52

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Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

WPI, CAS

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	EP 0 505 966 A1 (HOECHST AKTIENGESELLSCHAFT), 30 September 1992 (30.09.92), page 2, lines 1-3,38-40,48-51.	1,2
X	US 4 835 139 A (TICE et al.), 30 May 1989 (30.05.89), column 1, lines 50-63; column 3, line 65 - column 4, line 15; example 1; claim 1.	1
A	US 5 540 937 A (BILLOT et al.), 30 July 1996 (30.07.96), column 5, line 62 - column 6, line 32; example 3.	1,2,4
A	US 5 192 741 A (ORSOLINI et al.), 09 March 1995 (09.03.93), example 1; claim 1.	1,2,4
A	US 4 675 189 A (KENT et al.), 23 June 1987 (23.06.87), example 1; claim 1..	1,2,4
A	GB 2 165 517 A (DEBIOPHARM SA), 16 April 1986 (16.04.86), example 1.	1,2,4

Further documents are listed in the continuation of Box C.

See patent family annex.

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Date of the actual completion of the international search 27 September 1999 (27.09.99)	Date of mailing of the international search report 07 October 1999 (07.10.99)
Name and mailing address of the ISA/AT Austrian Patent Office Kohlmarkt 8-10; A-1014 Vienna Facsimile No. 1/53424/200	Authorized officer Mosser Telephone No. 1/53424/437

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La Recherchenbericht angeführtes Patentdokument Patent document cited in search report Document de brevet cité dans le rapport de recherche	Datum der Veröffentlichung Publication date Date de publication	Mitglied(er) der Patentfamilie Patent family member(s) Membre(s) de la famille de brevets	Datum der Veröffentlichung Publication date Date de publication
EP A1 505966	30-09-1992	AT E 124254 AU A1 13109/92 AU B2 653210 CA AA 2063883 CS A3 9200887 CY A 2032 CZ B6 284756 DE CO 59202649 DK T3 505966 EP B1 505966 ES T3 2076592 FI AO 921248 FI A 921248 FI B 99084 FI C 99084 HR A1 940838 IE B 67555 IL A0 101346 JP A2 5070363 NO A0 921147 NO A 921147 NO B1 302856 NZ A 242073 ZA A 9202130	15-07-1995 01-10-1992 22-09-1994 26-09-1992 14-10-1992 14-10-1992 20-02-1998 17-02-1999 03-08-1995 30-10-1995 28-06-1995 01-11-1995 23-03-1992 26-09-1992 30-06-1997 10-10-1997 26-09-1992 30-06-1997 17-04-1996 15-11-1992 23-03-1993 24-03-1993 28-09-1992 04-05-1998 26-08-1993 25-11-1992
US A 4835139	30-05-1989	CH A 661206	15-07-1987
US A 5540937	30-07-1996	AU A1 42022/93 AU B2 675788 CA AA 2100925 EP A1 585151 FR A1 2693905 FR B1 2693905 JP A2 6087758 NZ A 248207	10-02-1994 20-02-1997 28-01-1994 02-03-1994 28-01-1994 02-09-1994 29-03-1994 27-02-1996
US A 5192741	09-03-1993	AT A 2234/88 AT B 397035 AU A1 22326/88 AU B2 611944 BE AF 1001685 CA A1 1326438 CH A 6759468 DE A1 3822459 DE C2 3822459 DK A0 5189/88 DK A 5189/88 EEG AF 2009346 FI AO 884297 FI A 884297 FI B 96919 FI C 96919 FR A1 2620621 FR B1 2620621 GB A0 8722134 GB A1 2209937 GB B2 2209937 GR A 88100619 GR B 1002244 IE B 60608 IL A0 87790 IL A1 87790 IT A0 8805213 IT A 1225148 JP A2 1121222 JP B4 7013023 LU A 87340 NL A 8802323 NO A0 884154 NO A 884154 NO B 178604 NO C 178604 PT A 88557 PT B 88557 SE A0 8803321 SE A 8803321 SE C2 503406 LIS A 5776885 ZA A 8806827	15-06-1993 25-01-1994 23-03-1989 27-06-1991 06-02-1990 25-01-1994 30-11-1990 30-03-1989 07-07-1994 16-09-1988 22-03-1989 16-09-1989 19-09-1988 22-03-1989 14-06-1996 25-09-1996 24-03-1989 19-02-1993 28-10-1987 01-06-1989 03-07-1991 22-06-1989 22-04-1996 27-07-1994 31-03-1989 25-05-1992 20-09-1988 02-11-1990 12-05-1989 15-02-1995 06-04-1989 17-04-1989 19-09-1988 22-03-1989 22-01-1996 02-05-1996 01-10-1988 30-11-1992 20-09-1988 22-03-1989 10-06-1996 07-07-1998 30-05-1989

INTERNATIONAL SEARCH REPORT
Information on patent family members

International application No.

PCT/KR 99/00071

US A 4675189 23-06-1987

AT E	21624	15-09-1986
AU A1	77560/81	27-05-1982
AU B2	556754	20-11-1986
CA A1	1176565	23-10-1984
DE CO	3175230	02-10-1986
EP A2	52510	26-05-1982
EP A3	52510	08-09-1982
EP TD	52510	20-03-1986
EP B1	52510	27-08-1986
EP B2	52510	19-10-1994
HK A	204789	17-03-1989
IE B	52003	13-05-1987
IL AO	64298	28-02-1982
IL A1	64298	31-07-1985
JP A2	57118512	23-07-1982
JP B4	4040329	02-07-1992
MY A	835/87	31-12-1987
NZ A	198982	31-05-1985
PH A	19942	14-08-1986
SG A	944/87	06-05-1988
ZA A	8107973	27-07-1983

GB A 2165517

AT A	3011/85	15-10-1990
AT B	392586	25-04-1991
BE A1	903463	17-02-1986
CA A1	1259567	19-09-1989
CH A	660302	15-04-1987
DE A1	7536902	17-04-1986
DE C2	7536902	01-08-1991
DK AO	4745/85	16-10-1985
DK A	4745/85	18-04-1986
DK B	165355	16-11-1993
DK C	165355	05-04-1993
ES A1	547915	16-12-1986
ES A5	547915	24-12-1986
FR A1	8701495	01-03-1987
FR A1	2571617	18-04-1986
FR B1	2571617	19-08-1986
GB AO	8525398	20-11-1985
GBB A1	2165517	16-04-1986
GB B2	2165517	10-08-1988
IE B	58245	11-08-1993
IT AO	8505236	15-10-1985
IT A	1187226	16-12-1987
JP A2	61097216	15-05-1986
NL A	8502787	16-05-1986
NL B	192249	02-12-1996
NL C	192249	03-04-1997
SE AO	8504835	16-10-1985
SE A	8504835	18-04-1986
SE B	462780	03-09-1990
SE C	462780	10-01-1991
US A	4673595	16-06-1987